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Calprotectin Pegylation Enhanced Its Physical and Structural Properties

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Abstract Calprotectin is member of the S-100 protein family with a wide plethora of intra-and extracellular functions. Anticancer activities, antimicrobial effects and being a qualified disease marker are among the compelling features of this protein to be used as a pharmaceutical agent. However, there are several impediments to applications of protein pharmaceuticals including: proteolytic degradation, short circulating half-life, low solubility and immunogenicity. Pegylation is a common bioconjugation polymer capable of overcoming these drawbacks. Recombinant expression and purification of calprotectin along with its pegylation would result in enhanced pharmacodynamic and pharmacokinetic properties. Our florescence spectroscopy and far Ultraviolet-optical density results indicate that pegylation altered the physical and structural properties of the calprotectin to become in a more stable and functionally active state. Due to enhanced pharmacodynamic and pharmacokinetic properties of the calprotectin via pegylation, this study would pave the way for better in vitro and in vivo validations of calprotectin applications in medical practice.

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Abbreviations

CD	Circular dichroism
IPTG	Isopropyl-thio-β-D-galactoside
mPEG	Methoxy poly ethylene glychol
MRP	Myeloid inhibitory factor related protein
N_{i2}+-NTA	Nickel-nitrilotriacetic acid
PEG	Polyethylene glycol
SDS PAGE	Sodium dodecyl sulphate polyacrylamide gel electrophoresis

1 Introduction

Calprotectin is a calcium- and zinc binding protein of the S-100 protein family with a wide plethora of intra- and extracellular functions. It plays a pivotal role in the regulation of inflammatory processes and immune response. The protein complex is a 24 kDa heterodimer, incorporating a light (MRP8 also known as RP8/S100A8/L1L/P8) and a heavy (MRP14 MRP14 also known as S100A9/P14/L1H) chain. Calprotectin reported to have antimicrobial activities towards bacteria and fungi. The antimicrobial activity most likely is exerted via the ability of calprotectin to chelate Zn^{2+} which is essential for microbial growth. Moreover, it can induce cell death via autophagy and apoptosis; it have protective role in preventing exaggerated tissue damages; it acts as a potent amplifier of inflammation in autoimmunity as well as in cancer development and tumor spread, while regulates cell survival, neutrophil number and apoptosis [1–4].

The utilization of proteins and peptides as medicinal substances become to be an inevitable part of contemporary